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CASE REPORT

A NEW CASE OF INTRAGENIC DELETION OF THE *TCF4* GENE WITHOUT FEATURES OF PITT-HOPKINS SYNDROME

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Abstract: Different genomic alterations affecting the TCF4 gene are usually associated with Pitt-Hopkins syndrome (PTHS). This syndrome is a rare neurodevelopmental genetic disorder characterized by distinctive facial features, abnormal breathing, psychomotor delay and severe intellectual disability (ID). The genomic alterations include whole or partial gene deletion; balanced translocation disrupting the coding sequence of the gene; and intragenic variants. The TCF4 gene encodes a basic helix-loop-helix (bHLH) transcription factor 4. Using alternative promoters, TCF4 can be transcribed from a number of alternative initial exons, allowing for translation of variable protein isoforms containing different functional domains. Full-length TCF4 has two activation domains (AD1 and AD2) that are thought to modulate transcriptional activity, a NLS domain (nuclear localization signal) that controls subcellular localization and bHLH domain. Typical PTHS patients have aberration localized between exons 9 and 18 of the gene. On the other hand, variants affecting the first protein coding exons give rise to mild non-syndromic ID. We present a ten-year-old girl with psychomotor delay and mild ID without the typical features of PTHS. Genetic investigation using array-based comparative genomic hybridization, revealed a 73.45 kb deletion within the TCF4 gene. The deletion encompassing only exon 6 (NM_001083962). This deletion was not detected in both parents. Cytogenetic analysis excluded balanced translocation disrupting the coding sequence of the gene. To the best of our knowledge, this is the first case described in literature involving only exon 6. The findings in our patients support the notion that position of the alteration in TCF4 is relevant to the phenotype. Reporting our case we want to contribute to the phenotype-genotype correlation in patients with intragenomic deletion of TCF4 gene.

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CASE REPORT

Different genomic alterations affecting the TCF4 gene are usually associated with Pitt-Hopkins syndrome (PTHS). This syndrome is a rare autosomal dominant neurodevelopmental genetic disorder characterized by distinctive facial features, breathing abnormalities (hyperventilation episodes, apnea), ophthalmological disorders (strabismus, myopia), constipation, stereotypic movements, aggression, seizures, psychomotor delay and severe intellectual disability (ID).[1, 2] The genomic aberrations include whole or partial gene deletion, balanced translocation disrupting the coding sequence of the gene, and intragenic variants (frameshift, splice-site, nonsense or missense mutation, most localized between exons 7 to 19).[3, 4] In all PTHS patients with a heterozygous mutation in the *TCF4* gene whose parents were available for analysis, the mutation was shown to occur *de novo* supporting an autosomal dominant inheritance pattern secondary to haploinsufficiency of *TCF4*.[5]

The *TCF4* gene (OMIM *602272) is located on chromosome 18q21.1 and encodes a basic helix-loop-helix (bHLH) transcription factor 4. It can be transcribed using 21 mutually exclusive 5' initial exons (situated at various positions throughout exons 1-9), followed by constitutive internal exons 10-20, and 3' exon 21 (Figure 1). Full-length TCF4 has two activation domains (AD1 and AD2) that are thought to modulate transcriptional activity, a nuclear localization signal domain (NLS) that controls subcellular localization and a bHLH domain. The *TCF4* gene is highly expressed in

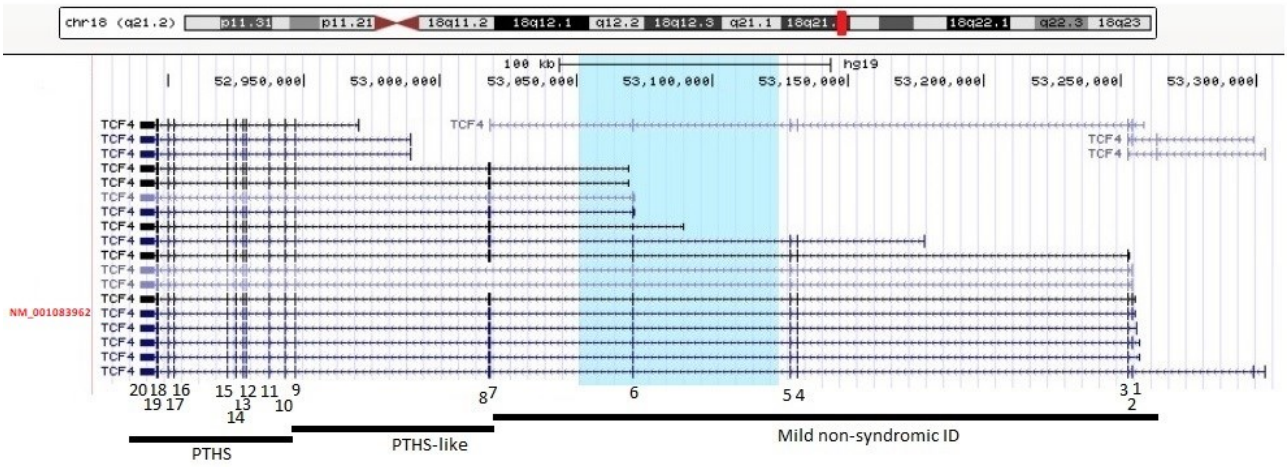

Figure 1: Screenshot from UCSC Genome Browser of *TCF4* genomic region (GRCh37/hg19). Exon numbering is referred to transcript NM_001083962. The blue shaded area indicates the deleted genomic region in presented case. The exons of the gene and predicted effect of variants affecting specific *TCF4* gene regions according to our patient and previously reported cases is shown at the bottom of the figure.

the developing and mature nervous system and interacts with other bHLH transcription factors important in nervous system development. Using alternative promoters, the *TCF4* gene can be transcribed from a number of alternative initial exons, allowing for the translation of variable protein isoforms containing different functional domains. Typical PTHS patients have an aberration localized between exons 9 and 18 of the gene. On the other hand, variants affecting the first protein-coding exons give rise to mild non-syndromic ID.[5]

We present unique partial deletion encompassing only exon 6 of the TCF4 gene identified by chromosomal microarray in a ten-year-old girl with strabismus, myopia, adiposity, developmental delay and mild ID without the typical features of PTHS. Chromosomal microarray was carried out using Agilent SurePrint G3 Unrestricted CGH ISCA v2 Human Genome 8x60 kit (Agilent Technologies, Santa Clara, CA, USA). Results were analysed using Agilent CytoGenomics (v4.0) software. Genetic testing revealed a de novo 73.45 kb heterozygous deletion in chromosomal region 18q21.1 spanning from 53,050,621 bp to 53,124,075 bp (GRCh37/hg19) within the *TCF4* gene. The deletion encompassed only exon 6 (numbering referred to transcript variant 1, NM_001083962) (Figure 1). The deletion was not detected in parents, and high resolution conventional cytogenetic analysis excluded balanced translocation disrupting the coding sequence of the gene.

The finding in our patient verifies the annotation that the position of the alteration in TCF4 is relevant to the phenotype. Most of the reported intragenomic deletions of the *TCF4* gene associated with PTHS include exons 9 to 18. Only several cases have been described with partial deletion of the *TCF4* gene affecting the first protein-coding exons.[2] Clinical presentation of these patients includes minor dysmorphic features, speech delay, mild to moderate ID, ophthalmologic anomalies and is not in the clinical score of PTHS.

Table 1. Genotype-phenotype correlation of published cases with deletion variants within the first eight exons of the *TFC4* gene

Patient	P29 [6]	P1 [6]	P4 [2]	P14 [2]	P19 [2]	P15 [2]	Present report
Deletion (exons)	4-6	4-6	4-5	4-8	4-8	5-6	6
Age at evaluation	3y 5m	55y	5y 4m	3y 3m	6y 5m	2y 11m	10y 2m
Intellectual disability	moderate to severe?	-	n.a.	n.a.	n.a.	n.a.	mild
Speech impairment	absent	-	babble 6m	babble 20m	-	babble 11m	+
Microcephaly	-	-	n.a.	n.a.	n.a.	n.a.	-
Motor incoordination	n.a.	-	+	+	n.a.	n.a.	+
Breathing anomalies	-	-	-	-	+	-	-
Constipation	n.a.	-	-	+	+	+	+
Brain abnormalities	n.a.	+	-	-	-	-	-
Ophthalmological anomalies	+	+	myopia strabismus	myopia strabismus	myopia strabismus	n.a.	myopia strabismus
Typical facial features	+/-	-	n.a.	n.a.	n.a.	n.a.	-

Legend: The exons numbering refers to transcript variant 1, NM_001083962; n.a. - not available

Our case further supports evidence that phenotype is largely dependent on the location of the alterations in the *TCF4* gene, with milder phenotypes caused by deletion affecting less important functional domains of the protein. [3, 6, 7] To the best of our knowledge, this is the first case described in the literature involving only exon 6 of the *TCF4* gene in a patient without features of PTHS. This case contributes to the genotype-phenotype correlation by which loss-of-function variants within exons 4-6 give rise to a phenotype with mild ID without typical features of PTHS.

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